

contacting said labeled high affinity TCRs with ligands;  
detecting the presence of the label thereby detecting the ligand to which the  
labeled high affinity TCR is bound wherein the high affinity TCR exhibits a  
dissociation constant for the ligand greater than about  $10^7$ .

26. The method of claim 25 where the high affinity TCR exhibits a dissociation constant for the ligand from about  $10^7$  to about  $10^{10}$ .
27. The method of claim 25 wherein the ligand is a peptide/MHC ligand.
28. The method of claim 27 wherein the peptide/MHC ligand is on the surface of a cell.
29. The method of claim 25 wherein the peptide is a superantigen.
30. The method of claim 25 wherein the label is selected from the group consisting of:  
fluorescent compounds, chemiluminescent compounds, radioisotopes and chromophores.
31. The method of claim 25 wherein the high affinity TCR carries one or more mutations in a CDR.
32. The method of claim 31 wherein the one or more mutations are in CDR3 $\alpha$  or CDR3 $\beta$ .
33. A method for using high affinity T Cell Receptors (TCRs) to detect ligands comprising the steps of:  
labeling high affinity TCRs;  
contacting said labeled TCRs with ligands;  
detecting the presence of the label thereby detecting the ligand to which the  
labeled TCR is bound wherein the high affinity TCR carries one or more  
mutations in a CDR.

34. The method of claim 33 wherein the one or more mutations are in CDR3 $\alpha$  or CDR3 $\beta$ .

35. The method of claim 33 wherein the ligand is a peptide/MHC ligand.

36. The method of claim 33 wherein the peptide/MHC ligand is on the surface of a cell.

37. The method of claim 33 wherein the label is selected from the group consisting of: fluorescent compounds, chemiluminescent compounds, radioisotopes and chromophores.

Sub 38. A soluble T cell receptor (TCR) having high affinity for a ligand exhibiting a dissociation constant for that ligand greater than about  $10^7$ .

39. The soluble T cell receptor of claim 38 wherein the TCR exhibits a dissociation constant for the ligand between about  $10^7$  to  $10^{10}$ .

Sub 40. The soluble TCR of claim 38 wherein the ligand is a peptide/MHC ligand.

41. The soluble TCR of claim 38 wherein the ligand is a superantigen.

42. The soluble TCR of claim 38 which is a mutant TCR carrying one or more mutations in a CDR.

43. The soluble TCR of claim 42 wherein the one or more mutations are in CDR3 $\alpha$  or CDR3 $\beta$ .

44. A soluble mutant T cell receptor (TCR) having high affinity for a ligand and having one or more mutations in a CDR.

45. The soluble TCR of claim 44 wherein the one or more mutations are in CDR3 $\alpha$  or CDR3 $\beta$ .

Sub 37 7 10  
46. The soluble TCR of claim 44 wherein the ligand is a peptide/MHC ligand.

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47. The soluble TCR of claim 44 wherein the ligand is a superantigen.

Sub B2  
48. A T cell expressing on its surface high affinity TCR's exhibiting a dissociation constant greater than  $10^7$  for a selected ligand.

49. The T cell of claim 48 wherein the TCR exhibits a dissociation constant between about  $10^7$  to  $10^{10}$  for said ligand.

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50. The T cell of claim 48 wherein the ligand is a peptide/MHC ligand.

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51. The T cell of claim 48 wherein the ligand is a superantigen.

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52. The T cell of claim 48 wherein the high affinity TCR is a mutant carrying one or more mutations in a CDR.

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53. The T cell of claim 48 wherein the high affinity TCR is a mutant carrying one or more mutations in CDR3 $\alpha$  or CDR3 $\beta$ .

Sub B3  
54. A T cell expressing on its surface a mutant high affinity TCR carrying one or more mutations in a CDR.

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55. The T cell of claim 54 wherein the high affinity TCR is a mutant carrying one or more mutations in CDR3 $\alpha$  or CDR3 $\beta$ .

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56. The T cell of claim 54 wherein the ligand is a peptide/MHC ligand.

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57. The T cell of claim 54 wherein the ligand is a superantigen.

58. A method for blocking autoimmune destruction of target cells comprising the step of contacting target cells with a soluble mutant TCR with high affinity for the site recognized by a T lymphocyte on the surface of the target cells, whereby the autoimmune destruction of the target cells is blocked, wherein the soluble, high affinity mutant TCR exhibits a dissociation constant for the ligand greater than about  $10^7$ .

59. The method of claim 58 wherein the high affinity mutant TCR exhibits a dissociation constant for the ligand between about  $10^7$  and  $10^{10}$ .

60. The method of claim 58 wherein the high affinity TCR is a mutant carrying one or more mutations in a CDR.

61. The method of claim 60 wherein the high affinity TCR is a mutant carrying one or more mutations in CDR3 $\alpha$  or CDR3 $\beta$ .

62. A method for blocking autoimmune destruction of target cells comprising the step of contacting target cells with a soluble mutant TCR with high affinity for the site recognized by a T lymphocyte on the surface of the target cells, whereby the autoimmune destruction of the target cells is blocked, wherein the soluble, high affinity mutant TCR carries one or more mutations in a CDR.

63. The method of claim 62 wherein the high affinity TCR is a mutant carrying one or more mutations in CDR3 $\alpha$  or CDR3 $\beta$ .

64. A method for using high affinity TCRs to kill an undesirable cell comprising the steps of:

coupling a TCR having a high affinity for cell surface marker of the cell with a therapeutic compound to form a therapeutic TCR derivative; and

contacting the therapeutic TCR derivative with the undesirable cell, wherein the high affinity TCR exhibits a dissociation constant for the cell surface marker of the undesirable cell greater than about  $10^7$ .

65. The method of claim 64 wherein the high affinity TCR exhibits a dissociation constant for the ligand between about  $10^7$  and  $10^{10}$ .

66. The method of claim 64 wherein the high affinity TCR is a mutant carrying one or more mutations in a CDR.

67. The method of claim 62 wherein the high affinity TCR is a mutant carrying one or mutations in CDR3 $\alpha$  or CDR3 $\beta$ .

68. A method for using high affinity TCRs to kill an undesirable cell comprising the steps of:

coupling a TCR having a high affinity for cell surface marker of the cell with a therapeutic compound to form a therapeutic TCR derivative; and  
contacting the therapeutic TCR derivative with the undesirable cell,  
wherein the high affinity TCR carries one or more mutations in a CDR.

69. The method of claim 68 wherein the high affinity TCR carries one or more mutations in CDR3 $\alpha$  or CDR3 $\beta$ .

70. A method of binding a high affinity TCR to a cell carrying a selected peptide/MHC ligand on the cell surface which comprising the steps of:

providing a mutant TCR exhibiting a dissociation constant of greater than about  $10^7$  for the selected peptide/MHC ligand;  
labeling the high affinity TCR;

contacting the labeled high affinity TCRs with a sample containing cells carrying one or more peptide/MHC ligands on the cell surface to bind the high affinity TCRs to selected peptide/MHC ligands present in the sample.

71. The method of claim 70 wherein the mutant TCR exhibits a dissociation constant between about  $10^7$  to  $10^{10}$  for the selected peptide/MHC ligand.
72. The method of claim 70 wherein the mutant TCR carries one or more mutations in CDR.
73. The method of claim 71 wherein the mutant TCR carries one or more mutations in CDR3 $\alpha$  or CDR3 $\beta$ .
74. A method of binding a high affinity TCR to a cell carrying a selected peptide/MHC ligand on the cell surface which comprising the steps of:

providing a mutant TCR having high affinity for the selected peptide/MHC complex and carrying one or more mutations in a CDR;  
labeling the high affinity TCR;  
contacting the labeled high affinity TCRs with a sample containing cells carrying one or more peptide/MHC ligands on the cell surface to bind the high affinity TCRs to selected peptide/MHC ligands present in the sample.

75. The method of claim 74 wherein the mutant TCR carries one or more mutations in CDR3 $\alpha$  or CDR3 $\beta$ .
76. A method for cloning the gene for a high affinity TCR mutant into a system that allows expression of the mutant on the surface of T cells comprising the steps of:

mutating TCRs to create high affinity TCR mutants which exhibit a dissociation constant for their cognate ligand of at least about  $10^7$ ;  
cloning said TCR mutants into a vector;  
transfecting the vector into T cells; and  
expressing the high affinity TCR mutant on the surface of T cells.

77. The method of claim 76, wherein the transfected T cells are used for recognition of selected peptide-bearing MHC cells.

78. The method of claim 76 wherein the high affinity TCR mutants carry one or more mutations in a CDR.

79. The method of claim 78 wherein the high affinity TCR mutants carry one or more mutations in CDR3 $\alpha$  or CDR3 $\beta$ .

80. A method for cloning the gene for a high affinity TCR mutant into a system that allows expression of the mutant on the surface of T cells comprising the steps of:

mutating TCRs to create high affinity TCR mutants carrying one or more mutations in a CDR;  
cloning said TCR mutants into a vector;  
transfecting the vector into T cells; and  
expressing the high affinity TCR mutant on the surface of T cells.

81. The method of claim 80, wherein the transfected T cells are used for recognition of selected peptide-bearing MHC cells.

82. The method of claim 80 wherein the high affinity TCR mutants carry one or more mutations in CDR3 $\alpha$  or CDR3 $\beta$ .

83. T cells made by the methods of claim 76.
84. A DNA sequence encoding a mutant high affinity TCR exhibiting a dissociation constant of greater than about  $10^7$  for its cognate ligand.
85. The DNA sequence of claim 84 wherein the TCR mutant exhibits a dissociation constant between about  $10^7$  and  $10^{10}$  for its cognate ligand.
86. The DNA sequence of claim 84 wherein the TCR mutant carries one or more mutations in a CDR.
87. The DNA sequence of claim 86 wherein the TCR mutant carries one or more mutations in CDR3 $\alpha$  or CDR3 $\beta$ .
88. A DNA sequence encoding a mutant high affinity TCR carrying one or more mutations in CDR.
89. The DNA sequence of claim 88 carrying one or mutations in CDR3 $\alpha$  or CDR3 $\beta$ .
90. A therapeutic TCR derivative which comprises a soluble high affinity single chain TCR coupled to a therapeutic compound.
91. The therapeutic TCR derivative of claim 90 wherein the therapeutic compound is an anticancer agent, a therapeutic radionuclide or a cytotoxic protein.
92. The therapeutic TCR derivative of claim 90 wherein the TCR specifically binds to a pathogen infected cell.
93. The therapeutic TCR derivative of claim 92 wherein the therapeutic compound is a molecule that is toxic to a pathogen.



94. The therapeutic TCR derivative of claim 90 wherein the high affinity TCR exhibits a dissociation constant greater than about  $10^7$  for its cognate ligand.
95. The therapeutic TCR derivative of claim 90 wherein the high affinity TCR carries one or more mutations in a CDR.
96. A method for treating disease in a patient comprising the steps of:
- removing wild-type T cells from the patient;
- transforming the T cells with the vector that expresses a high affinity TCR mutant, to express the high affinity TCR in the T cells;
- returning the transformed T cells to the patient;
- wherein the transformed T cells are activated to a greater extent than the wild type T cells of the patient.
97. The method of claim 96 wherein the high affinity TCR mutant exhibits a dissociation constant greater than  $10^7$  for a selected ligand.
98. The method of claim 96 wherein the high affinity TCR mutant carries one or more mutations in a CDR.
99. T cells made by the methods of claim 80.
100. A pharmaceutical composition comprising a high affinity TCR in a pharmaceutical carrier wherein the high affinity TCR exhibits a dissociation constant for a ligand of greater than about  $10^7$ .
101. A method of using the composition of claim 100 comprising administering the composition to a patient.

102. The method of claim 6, wherein said detecting step is performed by flow cytometry.

103. The method of claim 7, wherein said detecting step is performed by flow cytometry.

Accepted

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